International Research Journal of Pharmaceutical and Applied Sciences (IRJPAS)
Available online at **www.irjpas.com**
Int. Res J Pharm. App Sci., 2012; 2(5):139-142

Research Article

PHARMACOLOGICAL STUDY OF ANTI-DEPRESSANT LIKE ACTIVITY OF *CYPERUS SCARIOSUS OIL IN MICE*

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(Received: 18 September, 2012; Accepted: 29 September, 2012; Published: 29 October, 2012)
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Abstract: This study was planning to access the anti-depressant like activity of n-hexane extract of *Cyperus scariosus* oil in mice. Acute oral toxicity study was performed in mice. With two dose level at 100 mg/kg and 200 mg/kg anti-depressant activity was screened using Forced Swim Test(FST) and Tail Suspension Test(TST) in mice and result were compared with standard drug Imipramine (15 mg/kg,p.o). n-hexane extract of *Cyperus scariosus* oil significantly ($p<0.001$) reduced the immobility time in both dose level at FST and TST which is similar to standard drug imipramine. The result suggest the anti-depressant activity of n-hexane extract of *Cyperus scariosus* oil and may due to increase of norepinephrine level in synapses.

Key words: *Cyperus scariosus*, anti-depressant activity, Forced Swim Test(FST), Tail Suspension Test(TST), imipramine.

INTRODUCTION

Depression is considered as an affective disorder characterized by change in mood, lack of interest in the surroundings, psychomotor retardation and melancholia. The prevalence of depression in general population is estimated to be around 5%. At present 121 million people are estimated to suffer from depression. An estimated 5.8% of men and 9.5% of women experience a depressive episode in their lifetime with suicide being one of the most common outcomes of depression [1,2]. To date, the efficacy of the drugs for depression is very limited so the need for newer, better-tolerated and more efficacious treatments is remaining high. Therefore, herbal therapies should be considered as alternative/ complementary medicines. Recently, the search for novel pharmacotherapy from medicinal plants for psychiatric illnesses has progressed significantly.

Cyperus scariosus is known as Nagarmotha in India, is a plant of the Cyperaceae family. *Cyperus scariosus* is a delicate grass, available in different places of Bangladesh and in eastern and southern parts of Indo-Pak subcontinent [3]. Plant roots has a folkloric reputation as a cordial, tonic desicant, emmenagogue, diaphoretic and diuretic[4-5]. It remained to be an important ingredient of several prescriptions used in indigenous system of medicine to treat a variety of diseases including diarrhoea, epilepsy, fever, gonorrhoea, syphilis and liver damage [5]. The essential oil obtained on steam distillation of rhizomes and roots of the plant has its value in perfumary [6], and is also known to possess antibacterial [7] antifungal [8] as well as plant growth-regulating properties [9], analgesic and anti-diabetic activity[10], hepatoprotective activity[11] and hypotensive and spasmolytic activity[12].

Phytochemical studies revealed *Cyperus scariosus* oil mainly contains sesquinterpenes [13-14], and steroidal saponins[15].

Present study was aimed to screen anti-depressant activity of *Cyperus scariosus* oil in mice as currently a large number of oils are used in aromatheraphy to treat CNS disorders.

MATERIAL AND METHODS

Collection and authentication and extraction of drugs

The rhizomes roots and aerial parts of *C. scariosus* were collected from the CIMAP Farm, Lucknow and authenticated. A plant specimen was submitted in the Botanical and Taxonomic department of Institute (CIMAP, Lucknow) for identification and authentication. The dried rhizome and roots were powdered and extracted with n-hexane to obtain the oil.

Preliminary Physiochemical studies

The n-hexane extracted oil was tested for qualitative tests for oraganoleptic characters, solubility, specific gravity, refractive index, saponification value, iodine value and chemical tests for oils.

Drugs and Chemicals

The standard antidepressant drug imipramine (M/s. Alkem Ltd. Mumbai) was purchased from Retail Pharmacy; n-hexane was obtained from institutional store and was of analytical grade.

Animals

Inbred Swiss albino male mice (20-25 gm.) of were obtained from the animal house of Institute. The animals were maintained in a well-ventilated room with 12:12 hour light/dark cycle in polypropylene cages. Standard pellet feed (Hindustan Lever Limited, Bangalore) and drinking water was provided *ad libitum.* Animals were acclimatized to laboratory conditions one week prior to initiation of experiments. Before performing the experiment the ethical clearance was obtained from institutional animal ethics committee (IEAC).

Acute Oral toxicity Study

Acute oral toxicity study was carried out for n-hexane extracted *C. scariosus* oil using Acute Toxic Class Method as described in OECD (Organization of Economic Co-operation and Development) Guidelines No. 423. The oil was safe up to a dose of 2,000 mg/kg body weight.

Experimental design

The animals were divided into four groups of six animals each as follows:

Group I: Control: received 1 % aqueous solution of 2% Tween-80, p.o

Group II: Drug treated: received *C. scariosus* oil 100 mg/kg, p.o

Group III: Drug treated: received received *C. scariosus* oil 200 mg/kg, p.o

Group IV: Drug treated: imipramine15 mg/kg, p.o

Drugs were administered at 1ml/100 gm of body weight using intra-gestric feeding needle. Behavioural evaluation was carried out 60 minutes post drug/vehicle administration. The antidepressant activity of the test drug was evaluated using the following experimental models of depression Tail Suspension test (TST) and Forced Swim Test (FST).

Tail Suspension test (TST)[15, 17]

In tail suspension test the animals were hung by the tail on a plastic string 50 cm above the surface with the help of an adhesive tape, placed approximately 1 cm from the tip of the tail. Each animal under test was both acoustically and visually isolated from other animals during the test. The duration of immobility was observed for a period of 6 minutes. The duration of immobility was recorded during the last 4 minutes of the observation period. Mice were considered to be immobile only when they hung passively and were completely motionless. The test was conducted in a dim lighted room and each mouse was used only once in the test.

Forced swim test (FST)[16]

In forced swim test, each animal was placed individually in a glass chamber (25X 15 X 25 cm3) filled with water up to a height of 15 cm and maintained at 260C±10C. At this height of water, animals were not able to support themselves by touching the bottom or the sidewalls of the chamber with their hind-paws or tail. Water in the chamber was changed after subjecting each animal to FST because "used water" has been shown to alter the behavior. Animals were observed for duration of 6 minutes. The duration of immobility was recorded during the last 4 minutes of the observation period because each animal showed vigorous movement during initial 2 min period. The duration of the mouse was considered immobile when it floated motionlessly or made only those moments necessary to keep its head above the water surface. The water was changed after each test. The test was conducted in a dim lighted room and each mouse was used only once in the test.

Statistical analysis

Data are expressed as mean±SEM. The results were subjected to one-way analysis of variance (ANOVA), followed by Dunnet's multiple comparison test to compare the treatment groups with control group.

RESULT AND DISCUSSION

Acute Oral Toxicity Test

There was no mortality recorded at a dose of 2000 mg/kg of the extract. The LD50 of the Plant extract as per OECD guidelines falls under category 5 (LD50>2000 mg/kg).

Tail Suspension Test (TST)

Table-1: Effect of *C. scariosus* oil on immobility time in tail suspension test in mice.

Groups	Treatment	Dose (kg-1)	Immobility time (sec)
I	Control, 2% Tween-80	10 ml	196.45±5.4
II	*C. scariosus* oil	100 mg	143.45±3.8***
III	*C. scariosus* oil	200 mg	122.60±3.5***
IV	Imipramine	15 mg	101.40±4.5***

Values are mean ± SEM; for six animals in each group and statistical significance was calculated by ANOVA followed by Dunnett's test. ^{ns}-non significant, * $P< 0.05$; **P<0.01; *** $P< 0.001$ vs normal control.

After administration of a single oral dose, statistically significant decrease in the immobility time in TST was observed with drug treated animal at 100 and 200 mg/kg, when compared to the control group. Duration of immobility was significant at from the first day with standard drug imipramine. The extent of decrease in immobility time in was found at dose dependent and increases with the days of treatment. The results are shown in Table-1 and plotted in Fig-1.

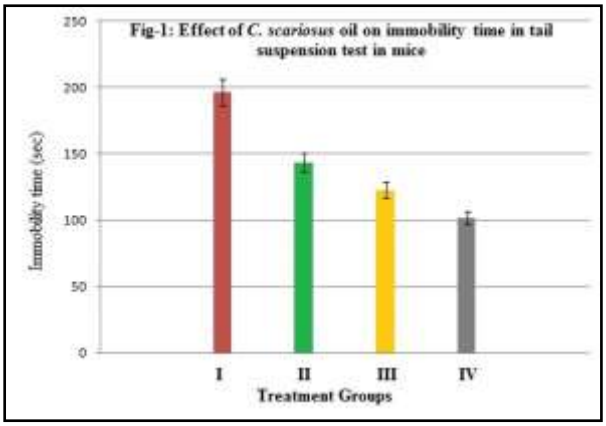

Fig-1: Effect of *C. scariosus* oil on immobility time in tail suspension test in mice.

Forced Swim Test (FST)

After administration of a single oral dose, statistically significant decrease in the immobility time in FST was observed with drug treated animal at 100 and 200 mg/kg, when compared to the control group. Duration of immobility was significant at from the first day with standard drug imipramine. The extent of decrease in immobility time in was found at dose dependent and increases with the days of treatment. The results are shown in Table-2 and plotted in Fig-2.

Table-2: Effect of *C. scariosus* oil on immobility time in Forced Swim test in mice.

Groups	Treatment	Dose (kg-1)	Immobility time (sec)
I	Control, 2% Tween-80	10 ml	206.50±6.8
II	*C. scariosus* oil	100 mg	153.62±4.2***
III	*C. scariosus* oil	200 mg	132.80±4.5***
IV	Imipramine	15 mg	111.63±3.6***

Values are mean ± SEM; for six animals in each group and statistical significance was calculated by ANOVA followed by Dunnett's test. ^{ns}-non significant, * P< 0.05; **P<0.01; *** P< 0.001 vs normal control.

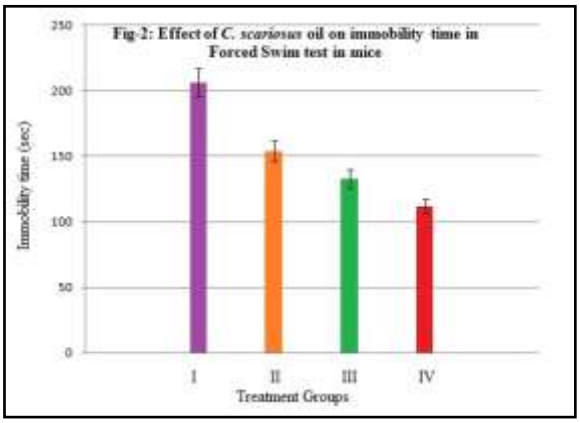

Fig-2: Effect of *C. scariosus* oil on immobility time in Forced Swim test in mice.

In the TST, mice are suspended by their tails for a defined period of time and their immobility is assessed. Acute administration of most antidepressants decreases immobility time in TST. The immobility exhibited by test animals in these models is indicative of a behavioral despair which reflects a depressive state[18].

Development of immobility when rodents are placed in an inescapable cylinder of water during FST reflects the cessation of their persistent escape-directed behavior. Conventional antidepressant drugs reliably decrease the duration of immobility in animals during these tests. This decrease in duration of immobility was considered to have a good predictive value in the evaluation of potential antidepressant agents.[19]

The precise mechanisms by which *C. scariosus* oil extract produced antidepressant like effect are not completely understood. However, according to our results, the pattern of behaviors exerted by the extract in the FST and TST is similar to those of imipramine which suggests that this plant extract acts probably by enhancement of norepinephrine neurotransmission as it is related to climbing behavior in the modified FST.

CONCLUSION

This study showed that *C. scariosus* oil possesses antidepressant effects. As the effect of extract was similar to that of imipramine, it may be concluded that this effect might be related to inhibition of norepinephrine uptake which eventually leads to increased availability of norepinephrine in synapses. Further research is underway.

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